

[0021] As used herein, the terms “nucleic acid,” “polynucleotide,” or “nucleic acid sequence” refer to a polymer of deoxyribonucleotides or ribonucleotides, in the form of a separate fragment or as a component of a larger construct. Polynucleotide or nucleic acid sequences of the invention include DNA, RNA, including mRNA and cDNA sequences.

[0022] As used herein, the term “polypeptide” refers to a polymer of amino acid residues in the form of a separate fragment or component of a larger construct. An example of a polypeptide includes amino acid sequences encoding a cytokine or fragments thereof. A polypeptide may encode for a functional protein or fragments of a protein. For example, an IL-4 polypeptide includes the full length protein sequence of IL-4 as well as fragments thereof consisting of a polymer of amino acids.

[0023] “Cytokine” as used herein means any number of factors that play a role in cellular regulation or differentiation. For example, cytokines can include the family of interleukins (IL) including IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-8, IL-9, IL-13, IL-14 as well as factors belonging to the transforming growth factor beta (TGF- $\beta$ ) superfamily, GM-CSF and interferon.

[0024] As used herein, the term “biological factor” means a number of factors that have biological activity or play a biological role. For example, biological factor includes polynucleotides, such as DNA, RNA, mRNA and cDNA, polypeptides, such as IL-4, IL-8, and IL-13 proteins and fragments thereof, as well as lipids such as cholesterol, fatty acids, and inflammatory mediators such as leukotrienes, prostaglandins and others.

[0025] The term “skin” means a tissue comprising a sheet of cells, one or several layers thick, organized above a basal lamina, and often specialized for mechanical protection or active transport. In a preferred embodiment, the skin is mammalian skin. In a more preferred embodiment the skin is human skin. The epidermis of the human skin comprises several distinct layers of skin tissue. The deepest layer is the stratum basalis layer, which consists of columnar cells. The overlying layer is the stratum spinosum, which is composed of polyhedral cells. Cells pushed up from the stratum spinosum are flattened and synthesize keratohyalin granules to form the stratum granulosum layer. As these cells move outward, they lose their nuclei, and the keratohyalin granules fuse and mingle with tonofibrils. This forms a clear layer called the

stratum lucidum. The cells of the stratum lucidum are closely packed. As the cells move up from the stratum lucidum, they become compressed into many layers of opaque squamae. These cells are all flattened remnants of cells that have become completely filled with keratin and have lost all other internal structure, including nuclei. These squamae constitute the outer layer of the epidermis, the stratum corneum. At the bottom of the stratum corneum, the cells are closely compacted and adhere to each other strongly, but higher in the stratum they become loosely packed, and eventually flake away at the surface.

[0026] The term "sample" refers to any preparation derived from skin of a subject. For example, a sample of cells obtained using the non-invasive method described above may be used to isolate polynucleotides, polypeptides, or lipids. In addition, the method of the invention can be used *in vitro*, for example with skin cells cultured on a solid or semi-solid support and organotypic skin constructs. In such instances, the skin cells may be from any source. A biological factor obtained from any *in vitro* or *in vivo* specimen, in purified or nonpurified form, can be used as the starting material for detection of a biologic activity, such as a dermatitis, provided it contains the biological factor of interest. For example, a sample may be used to detect a dermatitis by detecting polynucleotides, provided it contains, or is suspected of containing, the specific polynucleotide sequence encoding a polypeptide, such as a cytokine, which is indicative of a dermatitis.

[0027] Samples from a tissue may be isolated by any number of means well known in the art. Invasive methods for isolating a sample include the use of needles, for example during blood sampling, as well as biopsies of various tissues. Due to the invasive nature of these techniques there is an increased risk of mortality and morbidity. The present invention provides a method and kit useful for non-invasively obtaining a sample which may be used as a source for obtaining biological factors in the detection, diagnosis, or prognosis of various diseases, disorders, or inflammatory reactions. In a preferred embodiment the invention provides a non-invasive method for obtaining a skin sample for use in isolating biological factors, for example nucleic acids and/or polypeptides, to detect a dermatitis reaction. In this embodiment epidermal cells of the skin are scraped with a rigid instrument, for example a sterile #15 scalpel, however, it will be recognized that any number of rigid instruments capable of removing only the surface layer (*i.e.*,

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stratum corneum) of the skin may be used. Alternatively, instead of scraping the skin, the skin's epidermal layer may be removed by using an adhesive tape, for example, Duct tape (333 Duct tape, Nashua tape products) or Scotch® tape (3M Scotch 810, St. Paul, MN). However, a preferred method is to use D-SQUAME® (CuDerm, Dallas, TX) to strip the skin cell layer. In this embodiment the skin is stripped with the tape and the stripped cells and cellular material are then recovered from the scalpel, tape or other item. For example, tape used to obtain skin cells and cellular material may be centrifuged in a sterile microfuge tube containing lysis buffer. In the case of the scalpel the cells and cellular material may be transferred to a sterile petri dish and any cells present lysed therein with lysis buffer. The same lysis buffer may be reused for each piece of tape or scalpel used at a single skin site. For certain applications, the tape stripping method can be combined with the scraping method for removing cells and cellular material from the skin. The sample obtained may then be further processed, for example to isolate nucleic acids, polypeptides, or lipids. Preferably, the method utilized does not adversely affect the polynucleotide, polypeptide, or lipid level being measured. The invention provides, a rapid, non-invasive method for obtaining polynucleotides, such as mRNA, which are helpful to establish changes in the synthetic patterns of the skin's cells. The process of tape stripping itself has been shown not to affect the skin cytokine profile during the first few hours after the procedure is done. Using the scraping and stripping methods of the present invention the presence of a local or systemic disease, disorder, or inflammatory reaction may be identified, distinguished, or diagnosed, including genetic diseases. In the invention any reaction, disease, or disorder that corresponds to an induction of transcription and polypeptide synthesis may be detected by the methods of the invention.

**[0028]** Polynucleotides can be isolated from the lysed cells and cellular material by any number of means well known to those skilled in the art. For example, a number of commercial products are available for isolating polynucleotides, including but not limited to, TriReagent (Molecular Research Center, Inc, Cincinnati, OH ) may be used. The isolated polynucleotides can then be tested or assayed for particular nucleic acid sequences, including a polynucleotide encoding a cytokine.